Our Case No. 9793/115
Weickmann Ref. 11051P US-WO-2/WW
RDC Ref. RDID 0089 D US

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Application of:

Hans-Peter Josel et al.

Serial No. 09/801,157

Filing Date: March 7, 2001

For: Oligomeric Carrier Molecules with Defined Incorporated Marker Groups and Haptens

Examiner Jon D. Epperson, Ph.D.

Group Art Unit No. 1639

DECLARATION UNDER 37 CFR § 1.132

Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Dear Sir:

I, Milan Mrksich, declare the following:

1. My curriculum vita is attached.

2. I am currently appointed as Professor of Chemistry at the University of Chicago. I received a B.S. degree in Chemistry from the University of Illinois and a Ph.D. degree in Chemistry from the California Institute of Technology. I served as an American Cancer Society Postdoctoral Fellow at Harvard University before joining the Chemistry Faculty at the University of Chicago in 1996.

3. I direct a research group of approximately twenty students and postdoctoral fellows. My program is in the field of Chemical Biology and is broadly directed towards the development of molecular reagents and surface chemistries for studies in biochemistry and cell biology. I have more than fifteen years of research experience, including expertise in the following areas: organic synthesis, molecular biology, surface chemistry, biosensors, and protein biochemistry.

4. I am the coauthor of more than 100 peer-reviewed research publications and am an Inventor on 6 patents. I serve or have served as a Consultant to several

biotechnology companies, including IGEN, Cellomics, Surface Logix, ChemoCentryx, Helicos and WMR Biomedical. I serve as Vice-Chair of the Defense Sciences Research Council which is an advisory group to the Defense Advanced Research Projects Agency. Further, I serve as Chair of the Enabling Biotechnology Review Group that evaluates proposals for the National Institutes of Health, and on the Board of Governors for Argonne National Laboratory.

5. I have reviewed United States patent application Serial No. 09/801,157, the Office Action dated July 5, 2005 and the following references cited therein: Bredehorst, Brinkley, Merrifield and Massey.

6. United States patent application Serial No. 09/801,157 describes conjugates that are used in immunological assays. These assays take many forms, but in a common format the assay is used to analyze body fluids for antibodies that are produced in response to an infectious agent. Hence, the immunoassays are used, for example, to diagnose viral or bacterial infections in patients.

7. The claims relate to method of forming oligomeric molecules that are modified with haptens and either marker group(s) or immobilization group(s). The antigens are chosen so that they bind the antibody that is to be detected in a sample. The marker group(s) are chosen to permit observation of the oligomeric groups and the immobilization group(s) allow the oligomeric molecule to be attached to a solid phase. In one format, the antibody to be detected is incubated with an oligomeric reagent that both binds the antibody and carries a label. This mixture is applied to a solid phase that also binds the antibody to be detected. If the antibody target is present in the sample, it will bind to the solid phase and serve to bind the oligomeric reagent to the solid phase, where it can then be detected by way of the label.

8. Antigen reagents, such as those described above, have been prepared with a variety of methods. As the Applicants discuss, these reagents are often prepared by using synthetic polymers or biopolymers prepared with recombinant DNA methods. In both cases, the resulting reagents have limitations that can add to the cost and/or diminish the performance of the assay. Principal among these limitations is that it can be difficult to control the structures of the reagents. For example, the use of recombinant methods to prepare polypeptides is not effective for preparing many desired peptide sequences. In addition, the need to purify these reagents can lead to large variations in the properties of different batches.

9. The claimed invention recites a strategy that can prepare structurally well-defined oligomeric molecules, wherein the positions of the haptens, labels and immobilization groups can be defined precisely. In the language of the application, these sites are referred to as 'predetermined positions'. The method is based on using solid phase synthesis techniques in one of two strategies. In the first, amino acids that are tethered to the haptens, labels or

immobilization groups are used directly in the synthesis of the oligomeric molecules. In the second, amino acids that have protected amine or thiol side chains are used to prepare a oligomeric molecule. This molecule is then treated to remove the side chain protecting groups and reveal sites for the attachment of haptens and labels.

10. There are many benefits of using structurally well-defined and controlled oligomeric molecules. First, methods that give a single and well-defined molecule are more reliable in reproducibly preparing the material of interest. Second, the ability to control the absolute and relative positions of the haptens or labels serves to optimize their functions. For example, if two fluorescent labels are positioned close to one another, the fluorescent signal may be quenched. Hence, the ability to optimize the relative positions of the labels is important to providing a maximum signal. Third, since each oligomeric molecule is identical, they will each have identical properties, avoiding the distribution in properties that are common with heterogeneous reagents.

11. The examiner has rejected claims 33-39 under 35 USC § 103 as being unpatentable over the combined teachings of Bredehorst, Brinkley, Merrifield and Massey.

12. The examiner cites Bredehorst for teaching a method for the synthesis of a linear peptide carrier that contains one hapten molecule and multiple fluorescent labels. While this reference provides a clear example of the value of a single oligomeric molecule for use in immunoassays, it does not teach explicitly or give suggestions as to the use of solid phase syntheses to construct the molecule. Hence, I do not see in this reference information that is relevant to the claimed methods of preparing the oligomeric reagents. The examiner is aware of this deficiency but suggests that because the insulin carrier was purchased from Sigma, the "reference is silent as to whether or not the insulin was produced via solid-phase synthesis", leaving open the possibility that the cited example is relevant prior art to the claimed invention. I disagree with this interpretation. First, the authors of this reference purchased a reagent, and did not prepare that reagent using solid-phase synthetic methods. Second, a reading of this paper very strongly suggests that the authors did not appreciate that solid-phase synthesis of the carrier was important. For example, in comparing their method to alternate methods, they note that "polyamines such as polylysine and polyethyleneimine, which are available in degrees of polymerization up to several hundred, are suitable for simple reaction with FITC to provide multifluorophore labels", but go on to add that the close spacing of the fluorophores would lead to substantial quenching of the labels. They go on to say that the insulin carrier allows the conjugation of three fluorophores, and that "two of the fluorescein moieties are well removed from the DNP hapten, whereas the third is closer to the hapten and more susceptible to the known ability of DNP to quench the fluorescence emission..." This statement makes clear that the authors did not use insulin because it was an ideal carrier—since

one of the positions for fluorophore attachment was sufficiently close to the hapten to promote quenching—but rather because it was readily available. I see this as a clear example where had the authors appreciated that solid phase synthesis could be used to prepare the carrier, they would have done so, rather than settle for a commercially available carrier that was not optimal in its positioning of the labels relative to the hapten. Hence, I find that the Bredehorst reference provides information that speaks to the utility of oligomeric carrier molecules, but that it does not—either directly or indirectly—teach the use of solid-phase synthesis to prepare the oligomeric carrier molecules.

13. The examiner states that the combined teachings of Brinkley, Merrifield and Massey are prior art to claim 33. None of these references, however, refers to the solid-phase synthesis of a carrier from amino acids that are covalently bound to labels or haptens. Each of these references refers to the synthesis of peptides that contain amino acids that are not covalently bound to labels or haptens. Hence, the solid-phase synthesis does not follow the method described in claim 33.

14. In reference to claim 34, the examiner states that Merrifield shows an example of a solid-phase synthesis resulting in a peptide that contains a Cbzo protecting group. What the examiner does not state is that the purpose for using this protecting group is to prevent unwanted reactions that can occur during the synthesis of the peptide on solid-phase. The protecting group is not incorporated in order to permit the subsequent attachment of haptens or labels to the peptide. (Parenthetically, the examiner is not correct in noting that "the Cbzo protecting group is cleaved by base"; rather this protecting group is deprotected with acid).

15. In the same paragraph, the examiner states that Brinkley teaches the use of protecting groups, wherein "the N-terminal amino group is N-acylated [i.e., protected]". However, the examiner fails to recognize that this use of a protecting group is unrelated to the use of protecting groups in the claims of the application under review. In Brinkley, the acetyl protecting group serves to block the N-terminal amine from any subsequent chemical reactions. The acyl group cannot be removed—because any conditions that would result in hydrolysis of the acyl group would also result in hydrolytic cleavage of the amide bonds that constitute the backbone of the peptide carrier—and therefore is not useful for subsequent attachment of labels or haptens at the N-terminal amine group.

16. Brinkley provides a "brief survey of methods for preparing protein conjugates with dyes, haptens, and cross-linking reagents." It is significant that all of the examples in Brinkley involve the attachment of labels and haptens to peptide carriers that have already been formed, and does not discuss the direct incorporation of a label or hapten during the solid-phase synthesis. Further, Brinkley emphasizes methods that can be used to control the degree of labeling of a peptide carrier with labels or haptens. Brinkley recognizes that "when

proteins are labeled with fluorescent dyes, the fluorescence increases as more dyes are added; at the same time, however, the fluorescence efficiency decreases as a result of the quenching..." Brinkley goes on to describe strategies for optimizing the amount of dyes that are attached to each peptide carrier. This emphasis in the reference clearly teaches away from the concept of using a peptide carrier that has labels and haptens attached at predetermined positions. Further, I believe that one having ordinary skill in this art, at the time of the invention, would not recognize that solid-phase synthesis could be used to create carriers that had the optimal sites for attachment of the labels and haptens.

17. The examiner states that "it would have been *prima facie* obvious to one skilled in the art at the time (of) the invention to synthesize the peptide carrier molecule as disclosed by Bredehorst on a solid-support as disclosed by Merrifield because Merrifield explicitly states that his solid phase synthesis technology is ideally suited for peptide synthesis." I disagree because I believe that one skilled in the art at the time of the invention would not take away from the Bredehorst reference the insight to prepare the peptide carrier by solid-phase methods. Without this condition being met, the benefits of Merrifield's method to creating peptide carriers are not relevant.

18. The examiner goes on to claim that "a person of skill in the art would have been motivated to use the solid-phase synthesis technology disclosed by Merrifield to produce the insulin disclosed by Bredehorst because this technique overcomes prior difficulties with solubility and purification." The examiner's reasoning is faulty, because it suggests that a person of skill in the art was familiar with the strategy of using chemical synthesis of peptides to create the carrier molecules, but that the (then) conventional methods for peptide synthesis were too difficult to apply because of problems with solubility and purification. But just the opposite is true: a person of skill in the art was not familiar with the synthesis (using solid-phase methods or otherwise) of peptide carriers that had predetermined positions for attachment of labels and haptens. Hence, the motivation to use Merrifield's solid-phase technology is without basis.

19. In sum, the examiner has asserted one with ordinary skill in the art would have, at the time of the invention, recognized the combined teachings of the Bredehorst, Brinkley, Merrifield and Massey references. Notwithstanding my disagreement with several of the assertions made by the Examiner, I strongly disagree that one with ordinary skill in the art, at the time of the invention, would have been familiar with each of these references and would have recognized the combined teachings in the manner postulated by the Examiner. In hindsight, the Examiner's arguments can be interpreted in a manner that is consistent with the Office Action, but at the time of the invention, this argument calls for a level of insight and expertise that were very clearly beyond one of ordinary skill in the art.

of insight and expertise that were very clearly beyond one of ordinary skill in the art.

20. I further declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like are punishable by fine or imprisonment, or both, under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the above-referenced application or any patent issuing thereon.

Date: December 5, 2005 By: _____

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